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Short communication

Importance of tubule density to the fracture toughness of dentin

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ABSTRACT

Objective: The fracture toughness of dentin is critical to the prevention of tooth fracture. Within the tooth crown, the mechanical properties of dentin are influenced by spatial variations in the density and diameter of the dentin tubules with distance from the pulp. There are also relevant changes to the microstructure of dentin with age. In this investigation the importance of tubule density to the fracture toughness of dentin was evaluated in "young" and "old" age groups.
Methods: The variations in microstructure (density and diameter of tubules) from young and old donor teeth were studied by means of optical microscopy.
Results: A reduction in the density and diameter of tubules was identified to occur with aging. An approach previously proposed to study the mechanical behavior of porous materials was used to model the fracture toughness of coronal dentin in terms of the tubule characteristics. Results were then compared with published results from previous studies.
Conclusions: The model predictions were consistent with experimental results for the fracture toughness of dentin from young donor teeth, but overestimated the values that have been reported for "old" dentin.

1. Introduction

The "cracked tooth syndrome" was coined by Cameron (1964) over a half-century ago to describe the prominence of cusp fractures observed in restored teeth. Fracture is no less important to the field of restorative dentistry today (e.g. Barreto et al., 2015; Opdam et al., 2014).

The human dentition is subjected to a variety of cyclic stresses, including those of mastication and other *para*-functional activities. Acute stresses can promote the formation of microcracks in the tooth (Homewood, 1998), in addition to other events including trauma and restorative procedures (Bastone, Freer, & McNamara, 2000; Lee et al., 2014; Majd, Viray, Porter, Romberg, & Arola, 2012). Damage and microcracks in dentin may not cause fracture, but can facilitate the failure of restored teeth through fatigue crack growth (Arola, Huang, & Sultan, 1999; Bajaj, Sundaram, & Arola, 2008; Nalla, Imbeni et al., 2003). Spatial variations in the resistance to fracture of dentin can contribute to this process and is an important factor to consider.

Due to its importance, the fracture toughness of coronal dentin has been evaluated, with primary emphasis on the importance of tubule orientation. For instance, El Mowafy and Watts (1986) measured the fracture toughness of dentin for cracks oriented parallel to the dentinal tubules. The average fracture toughness reported was 3.08 MPa $m^{0.5}$, with no dependence of temperature between 0 °C and 60 °C. Imbeni, Nalla, Bosi, Kinney, and Ritchie (2003) employed a three-point bending approach to achieve crack extension perpendicular to the long axis of the tubules, and reported an average fracture toughness of 1.8 MPa $m^{0.5}$. Further, Iwamoto and Ruse (2003) measured the fracture toughness of dentin for three different orientations relative to the dentinal tubules, including directions denoted perpendicular, parallel, and parallel-transverse. No significant difference was found between the parallel (1.97 MPa $m^{0.5}$) and parallel-transverse (2.02 MPa $m^{0.5}$) orientations. However, the average value for the perpendicular orientation (1.13 MPa $m^{0.5}$) was significantly lower.

With the increase in senior dentate, aging has become of greater importance to the field of restorative dentistry (McNally, Matthews, Clovis, Brillant, & Filiaggi, 2014; Yellowitz & Schneiderman, 2014). Human teeth undergo changes with increasing age, including a decrease in the number of odontoblasts, an increase in dentin thickness and the formation of transparent dentin (Bernick & Nedelman, 1975; Murray, Stanley, Matthews, Sloan, & Smith, 2002; Nanci, 2012; Timiras, 2007; Toto, Kastelic, Duyvejonck, & Rapp, 1971). In addition, there are changes in mechanical properties, such as an increase in elastic modulus and hardness (Senawongse, Otsuki, Tagami, & Mjor, 2006), a decrease in strength

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(Arola & Reprogel, 2005; Kinney, Nalla, Pople, Breunig, & Ritchie, 2005) and decrease in fatigue crack growth resistance (Bajaj, Sundaram, Nazari, & Arola, 2006). There is also a reduction in fracture toughness with increasing age. Kinney et al. (2005) reported that the fracture toughness of transparent dentin, after nearly complete filling of the tubules by sclerosis, was 1.46 MPa $m^{0.5}$ and significantly lower than that for young dentin. The decrease in toughness appears to be independent of tubule orientation (Koester, Ager, & Ritchie, 2008b; Nazari, Bajaj, Zhang, Romberg, & Arola, 2009). Changes in the fracture toughness with aging may be related to dehydration (Toto et al., 1971), increase in the degree of mineralization (Porter et al., 2005), or an increase in the degree of crosslinking of the collagen fibrils (Miura et al., 2014). However, the most visible change is the decrease in diameter and density of the tubules, which results from an increase in mineral deposited within the tubules.

To the authors' knowledge, no investigation has examined the fracture toughness of coronal dentin in terms of tubule characteristics and number of obliterated tubules that occur with increasing age. Hence, the objective of this investigation was to examine the effect of tubule density on the fracture toughness of dentin from young and old donors and utilize an approach previously proposed for porous materials to quantify the spatial variations in fracture resistance.

2. Materials and methods

Human third molars were obtained from selected patients after written consent and following all the protocols required by the Dental Clinic at Universidad Cooperativa de Colombia (UCC). Exclusion criteria included presence of caries and previous restorations. The teeth were obtained from donors residing in Medellín, Colombia, and were divided into two age groups, namely a "young" group with donors between 18 and 25 years of age (N = 12), and an "old" group with donors between 47 and 65 years of age (N = 8). There were an equal number of male and female samples in both groups. Immediately after extraction, all the specimens were kept in Hank's Balanced Salt Solution (HBSS) at 2 °C to prevent dehydration (Habelitz, Marshall, Marshall, & Balooch, 2001). In addition, the specimens were tested within two weeks of extraction to limit the loss of mineral and potential degradation of organic materials.

Each molar was sectioned along its longitudinal axis (section A–A in Fig. 1a) using diamond abrasive slicing equipment with continuous water coolant. Secondary sections were made transversely (section A′–A′) in order to expose the dentin (Fig. 1b). For microscopic evaluations the specimens were embedded in cold-cured epoxy resin and then polished using silicon carbide abrasive paper with successively smaller particle sizes until reaching #1200 grit. Further polishing was then performed using diamond particle suspensions (3 μm particles) with standard red felt polishing cloth wheels. The polished specimens were then kept in a HBSS bath solution.

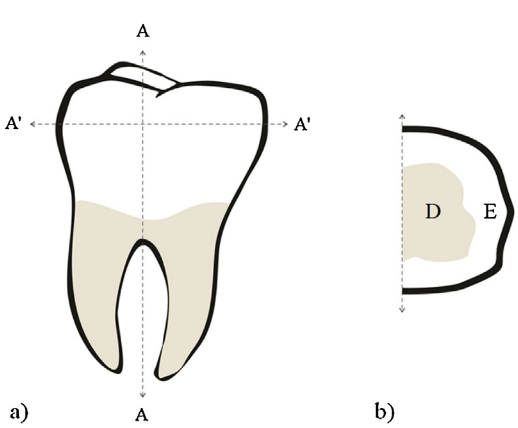

Fig. 1. Schematic diagram of a sectioned molar after (a) longitudinal (A–A), and (b) transverse (A′–A′) cutting. After sectioning the specimens, they were embedded in cold-cure epoxy resin with the sectioned surface facing outwards. The letters D and E refer to dentin and enamel, respectively.

The dentin sections were evaluated using optical microscopy (Axiovert 40 MAT, Carl Zeiss Microscopy, NY) to characterize the microstructure. Tubule density and tubule lumen diameter were measured within the regions corresponding to outer, middle and inner dentin. These measurements were located approximately at 0.5 mm, 2.0 mm and 3.5 mm away from the dentinal enamel junction (DEJ), respectively.

Measurement of the tubule density (ρ_t) and tubule lumen diameter (ϕ_l) was performed using commercial image analysis software (AxioVision LE). Seven randomly selected images with a constant area (approximate size of each image 80 μm × 100 μm) were obtained over the polished surface. The mean tubule diameter and number of tubules were calculated for each image. Values from the seven images were averaged and used to estimate the lumen area fraction (ξ) within the three regions of evaluation as

$$\xi = \frac{A_l}{A_T} \quad (1)$$

where A_l is the area (mm^2) occupied by lumens and A_T is the total area of dentin measured (mm^2) in each image. The average lumen area was calculated using the measures of tubule diameter and density.

3. Results

The microstructure of dentin from selected young and old donor teeth evaluated is shown in Fig. 2. Representative images are presented from the three regions of evaluation including the outer (Fig. 2a and b), middle (Fig. 2c and d) and inner (Fig. 2e and f) dentin. For both age groups the peritubular dentin can be seen surrounding each dentinal tubule. Several obliterated tubules are evident in the images for the old donor group, with greater number of filled tubules in the middle and outer regions. Micrographs of tubules within the outer dentin of young and old donor teeth obtained at higher magnification are shown in Fig. 3(a) and (b), respectively. An example of a tubule that has become filled with mineral is shown in Fig. 3(b). No obliterated tubules were evident in the dentin of the young donor group regardless of the region of evaluation.

Fig. 4 shows the estimates for the lumen area fractions (ξ) in the three regions of dentin evaluated. Overall, there was a significant decrease in ξ with proximity to the DEJ for both age groups ($p \leq 0.05$). In the young donor group the average ξ in the outer and inner regions was 3.7 ± 0.6% and 9.3 ± 1.0%, respectively. For the old group these values were 2.9 ± 0.4% and 9.6 ± 1.1%, respectively. The primary difference between the two age groups was the lower value of ξ (37% less) in the outer region of dentin for the old group. This difference is attributed to the reduction in diameter of the dentin tubules due to deposition of mineral within the lumens as a result of sclerosis. When comparing the results for ξ in each region between the young and old donors, significant differences were found only for outer dentin ($p \leq 0.05$).

4. Discussion

While the dentin tubules play an important role in tooth sensitivity and pain stimuli (Magloire et al., 2010), they also serve